Brucellosis in Ontario: A Case Control Study

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ABSTRACT

Data from cattle herds infected with brucellosis and from control (noninfected) herds were collected and analyzed using case control techniques.

It appeared that herds located close to other infected herds and those herds whose owners made frequent purchases of cattle had an increased risk of acquiring brucellosis, particularly those who made purchases from other herds or from cattle dealers. Infected herds had a lower level of vaccination than noninfected herds. However, the percentage vaccinated was highly variable in each group. Vaccination per se did not appear to adversely influence the interpretation of serological test results nor did it appear to protect the individual animal.

Once infected, the time required to become free of brucellosis was increased by large herd size and/or active abortion and/or loose housing. Closed herds also took longer to become brucellosis free than more open herds.

The percentage of animals removed from the herd was increased by active abortion. Those herds with multiple serological reactors (positives and questionables) at the first herd test after the imposition of quarantine had the highest percentage of cattle removed.

RÉSUMÉ

Cette étude visait à recueillir et à analyser, à l'aide de techniques axées sur l'éradication des cas, des données concernant des troupeaux de bovins atteints de brucellose et des troupeaux sains.

On réalisa que les troupeaux vivant à proximité d'autres troupeaux infectés et que ceux dont les propriétaires achetaient souvent des sujets, particulièrement d'autres troupeaux ou de commerçants, couraient un risque accru de contracter la brucellose. Les troupeaux infectés comptaient moins de sujets vaccines que les troupeaux sains. Le pourcentage d'animaux vaccinés variait cependant beaucoup dans les troupeaux infectés comme dans les sains. La vaccination per se ne semble pas nuire à l'interprétation des résultats d'épreuves sérologiques; elle ne sembla pas non plus protéger les animaux pris individuellement.

Une fois infecté, un troupeau demandait d'autant plus de temps pour devenir exempt de brucellose qu'il comptait un grand nombre de sujets et/ou qu'il connaissait des avortements et/ou qu'on y pratiquait la stabulation libre. Les troupeaux fermés prirent aussi plus de temps à devenir exempts de brucellose que les troupeaux plus ouverts.

Les avortements entraînaient une augmentation du pourcentage d'animaux qu'il fallait éliminer d'un troupeau. Ceux qui comptaient plusieurs réacteurs positifs et douteux, lors de la première épreuve sérologique ultérieure à l'imposition de la quarantaine, détenaient aussi le pourcentage le plus élevé d'animaux qu'il fallait éliminer.

INTRODUCTION

Brucellosis, infection with *Brucella abortus*, has been investigated in cattle in Canada since 1913 (15). In recent years the level of this disease has undergone a continual decline as demonstrated by (a) reduction in suspect Milk Ring Tests (MRT) from 2.6% in 1960 to 0.04% in 1971 and (b) a reduction in suspect Market Cow Tests (MCT) from 2.8% in 1964 to 0.7% in 1973 (2). Concomitantly, calf-

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hood vaccinations have shown a continuous decline from a peak of approximately 170,000 in 1968 to 12,000 by 1973 (24) under federal and provincial endorsement in Ontario.

In spite of over 60 years of continual research there remains a paucity of objective guidelines for the control and eradication of brucellosis. This deficit of knowledge together with the recent dramatic increase in apparent prevalence of infection (2), particularly in eastern Ontario, demonstrates the need to establish new guidelines for the use of the regulatory veterinarian.

The objective of this study was to utilize the basic data, collected by the district veterinarian, Health of Animals Branch (HofA), Agriculture Canada in the course of his investigation, to assess associations of possible importance in the epidemiology of this disease.

MATERIALS AND METHODS

Data were collected for a number of variables in this study. The variables listed in Table I were selected after reviewing the available literature for variables of potential importance in the epidemiology of brucellosis. Further criteria of direct importance to the HofA are listed in Table II.

Study Area

Peterborough and Victoria counties were selected as the study area because they had a relatively high prevalence of brucellosis (2), a necessity in the performance of a case control study (20) and were assessed by federal authorities as approximating counties of eastern Ontario. Situated in south-central Ontario as depicted in Figure 1, these continguous counties are comprised of small farms averaging 38 head (2) of predominantly beef cattle in an area that has resisted the trend toward the abandonment of the family farm system of agriculture (17). Average annual precipitation is 32 inches (8) with maximum daily temperatures varying from -5°C to 0°C in January and from 25°C to 30°C in July.

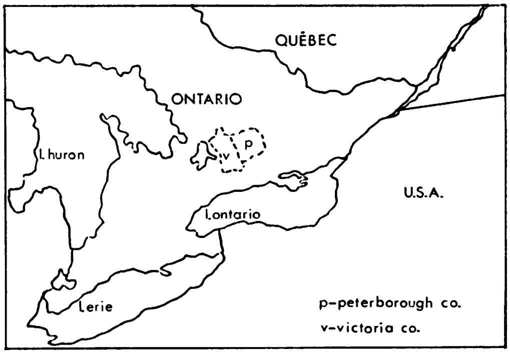

Fig. 1. Map of southern Ontario depicting location of Peterborough and Victoria counties.

These counties have been of Brucellosis Certified Area status (BCA) since 1961 (1) with the most recent recertification (statutory 18-month periods of testing ending March 1, 1970 for Victoria County and August 2, 1971 for Peterborough County) based on internationally used testing procedures (MRT, MCT and herd tests). On the basis of these measures, federal authorities considered that neither of the two counties contained more than (a) 1% of the herds or one herd, whichever was the greater, infected with brucellosis during the 18-month period, (b) 2% of the cattle infected during that time and that (c) brucellosis had been eradicated in all herds in which it was previously known to exist (1). The period of our study dated from the termination of the respective recertification periods to April 1975.

Farm Selection

The cooperation of all levels of the HofA, particularly the district veterinarian, made all herd records available. All herds designated since recertification as infected on the basis of bacteriological or serological (the tube agglutination test is standard in Canada) evidence of brucellosis were selected as cases (N=46).

From the complete list of all county herds (cases excluded) control herds were randomly selected employing a table of random digits (30) at a ratio exceeding 2:1 with respect to the number of cases. This number was later reduced to 86 viable herds by deletion of farm operations inactive over the period of study.

DATA COLLECTION

Data were collected from all farms using a mailed questionnaire. An evaluation of the clarity of the questionnaire and its acceptance at the farm level was conducted with a pilot mailing to a randomly chosen list of farmers in another Ontario county. The quantity and quality of the responses derived served as guidelines for the final questionnaire. The subsequent mailing of the questionnaire to the owners of the herds under study followed by two mailed and one verbal reminder at biweekly intervals yielded 80% and 72% viable responses from cases and controls respectively. Additional data from cases were obtained from HofA records.

TABLE I. A Summary of Factors of Importance in the Epidemiology of Brucellosis

Variable	Summary Statements	Ref. No.
1. Herd size	— Higher proportion infected in large herds	9
	— Difficult to clean up large herds	28
	— Multiple reactors occur in larger herds	22
2. Registration status	— Higher percentage of animals infected in purebred beef herds than grade herds	22
3. History of previous reactor(s)	— No such history in herds with 1 and 2 reactors in 1968 testing of certified-free areas in U.S.A.	18
4. Exposure	— Tendency to spread to adjacent premises	23
a) on pasture	— Contiguity to infected herds frequently associated with infection in new herds	34
b) on purchase	— Infection in big majority of cases acquired from outside source	9
	— Even calf purchases dangerous	13
	— Percentage infected cattle higher where more interchange of cattle occurs	14
	— Intensive market trade fatal to disease control	33
	— Incidence highest in areas of habitual movement of dairy cattle	12
	— Closer control of markets should be attempted	16
5. Proximity to infected herds	— See 4(a) above	
6. Stabling	— Yard and parlour system does little to limit spread of brucellosis	25
	— Trend to self-feeding silage unit increases disease eradication problems	23
7. Vaccination level	— Herd vaccination status did not significantly reduce herd infection rate	19
	a) higher herd percentage vaccination gives higher individual protection	27
	b) in a beef area with 27% overall vaccination: vaccination not a prime cause of suspect reaction	27
	— In an area of overall 80% vaccination individual less likely to be reactor	27
8. Farm density	— Infection higher in areas of greater cattle concentration	12
9. Use of maternity pens	— Infection higher in areas of greater cattle concentration	12
	— A major eradication problem is convincing farmers to use isolation at calving	23
	— Regulation made compulsory housing of any animal which has retained fetal membranes	33
	— Little thought given by farmers to providing adequate calving pens	4
10. Insemination methods	— Although bull not regarded as a major source of infection, the disease can be spread by AI if semen infected	29
	— Natural service is not a major factor in spread in a herd	26
	— Infected bull would not spread infection naturally, would when semen used artificially	21
11. Herd type	— Greater incidence in dairy herds could be due to greater interchange of stock	9

Data Organization and Analysis

The temporal and spatial distribution of cases were investigated. In addition, most data were transferred to computer cards in a form suitable for statistical analysis. This necessitated the assignment of numerical values to many variables (Tables III and IV).

The analyses of a case control study is designed to ascertain if significant differences exist between the case group and the control group with respect to the relative frequency of one or more variables. When more than one variable is used the analytic technique must take into account not only the differences between the groups for each variable but also the correlation(s) between the variables. Discriminant analysis (30) which is essentially a multivariate t test has this ability and was used in this study for the analysis of difference between the groups.

In addition, discriminant analysis was used to test for differences among groups of cases after all of the cases had been divided into four groups on the basis of the TAT results at the first herd test following the imposition of quarantine.

Stepwise regression analysis (10) was also applied to the data from case herds in an attempt at predicting (a) the number of months, (b) the number of tests required to remove infection from a given herd and (c) the percentage of the herd removed as reactors during the investigation.

Finally, the relationship between vaccination status and serological reactions was assessed using chi-square analysis (31).

RESULTS

Figure 2 graphically displays the increased incidence of brucellosis which began in 1973. Table V depicts the deviation (in the form of clumping) from the expected Poisson distribution for both cases and con-

TABLE II. Variables, Related to Brucellosis Quarantine, of Importance to Health of Animals Personnel

Variable	Department Interest
12. Diagnostic Test	Comparative assessment of outcome of various tests
13. Husbandry	Assessment of its importance in an infected herd
14. Months to quarantine release	Spread of infection from infected premises. Manpower and costs
15. Tests to quarantine release	Spread of infection from infected premises. Manpower and costs
16. Percentage removals from herd	Prediction of budgetary requirements

TABLE III. Coding for Variables Employed in Discriminant and Stepwise Regression Analyses

Variable	Code
1. Herd Size	Actual number (no.)
2. Herd registration status	PB = 1 Grade = 2
3. History of reactor (since 1959)	Yes = 1 No = 2
4. Exposure (replacement and/or pasture)	Nil = 1, Light = 2, Moderate = 3, Heavy = 4
5. Proximity to infected herd	Actual no. of infected herds within 2 miles
6. Stabling	Tied = 1 Loose = 2
7. Vaccination level of cattle tested	Actual percent vaccinated
8. Farm density of area	Actual no. of controls within 2 miles
9. Maternity pen usage	Yes = 1, Sometimes = 2, No = 3
10. Breeding method	AI = 1, Both = 2, Nat = 3
11. Herd type	Dairy = 1, Mixed = 2, Beef = 3
12. Diagnostic test	Abortion = 1 MCT = 2 MRT = 3 Traceback = 4 Test area = 5
13. Husbandry	Excellent = 1 Good = 2 Fair = 3 Poor = 4
14. Number of tests to quarantine release	Actual number
15. Number of months to quarantine release	Actual number
16. Percent removed of those tested	Actual number

TABLE IV. Codes for Exposure Variables Employed in a Discriminant Analysis of Brucellosis Case and Control Herds in Two Ontario Counties

(A)	Assignment of Ratings Exposure Method		Rating[a]
	Pasture with other cattle		4
	Pasture adjacent to other cattle		2
	Natural increase		0
	Purchase whole herd		1
	Purchase from dealer		6
	Purchase direct from herd		2
	Community sales purchase		6
	Purebred and production sales purchase		1
	Lease		2
(B)	Assignment of Codes Total of Herd Ratings	Subjective Evaluation	Code
	0	Nil	1
	1 — 3	Light	2
	4 — 5	Moderate	3
	6 +	Heavy	4

[a]Ratings were based on authors' experience and information from available literature

trols. The coefficient of dispersion (1.96) for cases exceeded that for controls (1.40).

Discriminant analysis significantly (F statistic significan at $p \lesssim 0.05$) differentiated between the case and control herds using the variables shown in Table VI. After the first six variables had entered the discriminant function, 100 of 132 herds (30 of 46 cases and 70 of 86 controls) were correctly categorized. The addition of further variables did not alter this categorization. The differences between the case and control farms with respect to purchasing practices are shown in Table VII (F statistic significant $p \lesssim 0.05$).

Discriminant analysis using four factors (no. tests, no. months, % removals and total removals) was unable to differentiate between case herds having only negative, questionable or positive serological test results (Table VIII). However, these three groups were significantly different ($p \lesssim 0.05$) from the group having both positive and questionable test results.

With variables 1-11 (Table III) and variables 12 and 13 (Table IV) as independent variables in stepwise regression analyses, the amount of variation in the dependent variable accounted for was 0.49 with number of months required to obtain "clean status" as the dependent variable (Table IX), 0.23 with number of tests required to obtain "clean status" as the dependent variable (Table X) and 0.31 with percent removals from the herd as the dependent variable (Table XI).

The association between vaccination status and serological test result(s) was not significant ($p > 0.05$). The data are shown in Table XII.

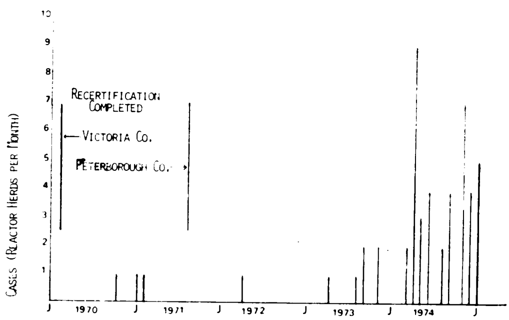

Fig. 2. Temporal distribution of cases of brucellosis in two eastern Ontario counties.

DISCUSSION

Before considering specific results, it seems prudent to discuss the methods of data collection and analysis and the possible effects this had on the results. First, data were collected from all farms using a mailed questionnaire. This technique has recently been evaluated (11) for data collection on calf diseases and has been found acceptable. The acceptability and clarity of the questionnaire was assessed in a pilot study prior to its general usage. Further, we evaluated the rate of return for the total questionnaire and the response rate for particular questions and found equivalent rates for the cases and controls.

Data analyses were primarily conducted using two multivariate statistical techniques, discriminant analysis and multiple stepwise regression. These techniques assume continuous data and treat all variables as such. Our inclusion of a number of attribute type variables (following coding) to a certain extent detracts from the overall usefulness of the analyses. However, we and others (30) feel the procedure is justified since we are not reaching definitive conclusions, but rather are attempting to identify important factors for further study. In addition, our analyses were probably less efficient and more conservative because no attempt was made to normalize the data or ensure homoscedasticity. When data were missing (e.g. failure to reply to certain questions) average values for the respective counties were used.

TABLE V. Results of Poisson Analysis of Spatial Distribution of Case and Control Herds in Two Ontario Counties

Farms Per 4 Miles Square (X)	Frequency (Y)	Expected Frequency (Y)
(A) Cases (N = 46)		
0	176	164
1	18	36.7
2	8	4.1
3	1	0.3
4	1	0.0
5	1	0.0
	205	
	Chi-Square = 4.44[a]	
	Coefficient of Dispersion = 1.96	
(B) Controls (N = 86)		
0	146	134.9
1	38	56.6
2	17	11.9
3	2	1.7
4	2	0.2
5	0	0.0
	205	
	Chi-Square = 10.78[a]	
	Coefficient of Dispersion = 1.40	

[a]Significant at $p \leq 0.05$

The marked increase in the incidence of brucellosis in these counties occurred in other counties and provinces and was a major reason for initiating this study. The spatial clumping of farms which was noted in this study was not surprising given the topographic characteristics of these counties (6, 7). The larger size of the coefficient of dispersion for cases could be interpreted as resulting from an additional clumping effect exerted by the contagious nature of this disease.

When this study was initiated it was deemed important to assess possible reasons for a herd becoming infected. In addition, a detailed survey of the case records indicated that for at least 48% of the case herds no source of infection had been determined. While we are still unable to state the source of infection for any given farm there were marked differences between the case and control herds which are suggestive of important sources.

The variable "number of infected herds within two miles" was the best statistical discriminator (Table VI), reemphasizing the contagious nature of this infection and reflecting the additional clumping of cases noted in the Poisson analysis (Table V). Although there was an extensive degree of pasturing with or adjacent to other herds (overall 82%), pasturing by itself was not a good discriminator. This suggests that pasturing with other cattle was

TABLE VI. Results of Discriminant Analysis Between Case and Control Herds in Two Ontario Counties

Step No.[a]	Variable[b]	Cases (N = 46) Mean ± SD	Controls (N = 86) Mean ± SD	Overall Mean
1	No. reactor herds within 2 miles	1.35 ± 1.4	0.43 ± 0.6	0.75
2	Exposure rating (Contact with other cattle)	3.28 ± 0.9	2.77 ± 1.0	2.95
3	Herd type	2.41 ± 0.9	2.62 ± 0.7	2.55
4	Percent susceptibles vaccinated	16.50 ± 21.7	26.77 ± 27.3	23.19
5	History of previous reactor	1.67 ± 0.5	1.60 ± 0.5	1.63
6	Stabling type	1.65 ± 0.5	1.51 ± 0.5	1.56
7	Herd registration	1.98 ± 0.1	1.90 ± 0.3	1.92
8	Herd size	59.80 ± 40.5	50.27 ± 43.8	53.59
9	Control herds within 2 miles	0.89 ± 1.1	0.85 ± 0.9	0.86
10	Use of maternity pens	2.13 ± 0.5	2.20 ± 0.8	2.17
11	Insemination method	2.35 ± 0.8	2.41 ± 0.8	2.39

[a]Refers to step at which variable entered the discriminant function
[b]For coding variables, see Table III

TABLE VII. Results of Discriminant Analysis of Variables Relating Stock Replacements in a Case Control Study of Brucellosis in Two Ontario Counties

Step No.[a]	Variable[b]	Cases (N = 46) Mean ± SD	Controls (N = 86) Mean ± SD	Overall Mean
1	Purchases direct from another herd	0.43 ± 0.5	0.21 ± 0.4	0.29
2	Replacements from natural increase	0.74 ± 0.4	0.90 ± 0.3	0.84
3	Purchases from livestock dealer(s)	0.17 ± 0.4	0.05 ± 0.2	0.09
4	Purchase of an entire herd	0.04 ± 0.2	0.01 ± 0.1	0.02
5	Purchase from community sale(s)	0.26 ± 0.4	0.23 ± 0.4	0.24
6	Purchase from purebred or other production sale	0.11 ± 0.3	0.10 − 0.3	0.11

[a]Refers to step at which variable entered the discriminant function
[b]Variables coded as 1 if used, 0 if not used

TABLE VIII. Results of Discriminant Analysis Amongst Cases in Two Eastern Ontario Counties

		Herds Having					
		Negatives Only	Questionables Only	Positive(s) Only		Positives and Questionables	
Step No.[a]	Variable	Mean ± SD	Mean ± SD	Mean ± SD	Mean of Three Groups	Mean ± SD	Overall Mean
1	No. removed	3.26 ± 4.6	4.39 ± 4.6	8.00 ± 6.3	4.4	19.3 ± 7.1	5.37
2	Percent removed	15.05 ± 26.9	19.67 ± 26.8	47.0 ± 44.2	21.4	41.33 ± 52.0	22.74
3	No. of tests to clean status	3.42 ± 0.7	3.89 ± 0.8	4.00 ± 0.9	3.7	3.33 ± 0.6	3.67
4	No. of months to clean status	7.03 ± 2.3	9.33 ± 4.4	6.05 ± 2.9	7.9	12.67 ± 5.5	8.17

[a]Refers to step at which variable entered the discriminant function

TABLE IX. Results of Stepwise Regression Analysis[a] for Case Herds in Two Ontario Counties

Variable Entered	Correlation Coefficient	Step at Which Variables Included	F Value to Enter Regression	Cumulative R-Squared
Herd size	0.416[b]	1	9.19[b]	0.17
Exposure	−0.189	2	4.28[b]	0.25
Test (diagnostic)	−0.208	3	4.33[b]	0.32
Registration status	0.164	4	6.67[b]	0.41
Vaccination level	0.010	5	2.46	0.45
Insemination method	0.194	6	3.67[b]	0.49

Total $R^2 = 0.52$
[a]Dependent variable = No. of months to clean herd status. First six independent variables entered into the regression are shown. Independent variables No. 1 — 13 Table III
[b]Denotes significance at $p \lesssim 0.05$

TABLE X. Results of Stepwise Regression Analysis[a] for Case Herds in Two Ontario Counties

Variable Entered	Correlation Coefficient	Step at Which Variables Included	F Value to Enter Regression	Cumulative R-Squared
Stabling method	0.350[b]	1	6.16[b]	0.12
Exposure	−0.281	2	4.56[b]	0.21
Husbandry	0.213	3	0.64	0.22
Herd size	0.128	4	0.63	0.23

Total $R^2 = 0.26$
[a]Dependent variable = No. of tests to clean herd status. First four independent variables entered into regression are shown. Independent variables No. 1 — 13 Table III.
[b]Denotes significance at $p \lesssim 0.05$

only important if an infected herd was located within two miles. Some outbreaks were traced directly to pasture contact.

Exposure, i.e. a measure of outside herd contact, was the second variable used for discrimination, with a higher exposure rating in the case herds, revealing an above average tendency towards openness. This concurs with Stuart's observations in dairy cattle in California (32). A separate, more detailed analysis of exposure (Table VII) indicated that in general, owners of case herds made more frequent purchases.

Relative to control herd purchases, case herds purchased twice as often from another herd, three times as frequently from livestock dealers and four times as frequently by purchasing an entire herd. The most frequent source of purchased animals were first from another herd, second from community sales, third from a purebred sale and fourth from livestock dealers. Very few farmers purchased entire herds. Bearing in mind the absolute number of purchases and the relative differences between the case and control herds, purchasing directly from other herds or via a livestock dealer appear to be the most important possible sources of infection in purchased cattle. Although community sales and purebred sales do not appear to be important sources of infected animals the over reliance of the case herds on purchases in general is noteworthy. Numerous authors have associated extensive cattle movement with the spread of brucellosis (9, 12, 13, 14, 33) and in our opinion the most organized approach to solving this problem is the restocking scheme of the British Veterinary Association (5).

Reactor herds tended to be of dairy type, in agreement with the observations of Christie (9). Perhaps the necessity of production maintenance requires more frequent purchases in dairy herds, exposing them to the risks previously described.

The overall vaccination level of susceptible animals in these counties was 23% (Table VI). Although a large difference existed between the level in cases (16%) and controls (26%) the amount of variability within each group reduced its value as a discriminator. Other studies in Ontario[1] also failed to detect significant differences in vaccination level between infected and possibly noninfected herds.

Control herds had a more frequent history of previous reactors than cases. This could indicate that department policy (1) in dealing with infected herds was effective and in addition, once experiencing an outbreak of brucellosis in his herd, a farmer might beneficially modify his management practices to lower the likelihood of reinfection (e.g. purchase less frequently).

Loose stabling was more frequent in case herds. However, we cannot find a logical reason for this differentiation. It is possible that exposure, vaccination status, stabling type and other variables do not describe their specific effects *per se* but rather a general attitude of owners towards control of this and other diseases.

Our attempt to discriminate between the groups of case herds classified according to the serological results at the first test following the imposition of quarantine was made in order to predict the outcome of brucellosis in that herd. In other words, given that both positive and questionable animals are found in the herd, is the outcome different than if only questionable animals are present? The categories were arranged from low (negative test results) to high (positive and questionable results) factor values. The number of cattle removed was the best discriminator ($P \lesssim 0.05$) and exhibited an increasing linear trend as the test results went from low to high. This suggests that the results of this test are useful in predicting the relative numbers of animals which will eventually be removed from that herd.

Further attempts at predicting the outcome of brucellosis were made using stepwise regression analysis. This technique was selected since the dependent variable (no. tests, no. months and % removals) in each analysis was quantitative and at least semicontinuous. Regression analyses of the number of months and number of tests were arbitrarily separated since the number of months often reflects repeated individual tests on animals with questionable reactions, whereas the number of tests represent complete herd tests.

Although multiple regression techniques do not lead to unique solutions we infer from the results of regression and from the correlation coefficients that:

1. Larger herds require more time (no. tests and no. months) to clean up, sup-

[1]Martin, S. W. Data presented at HofA Seminar on Brucellosis, Ottawa, Ontario, March 1975.

TABLE XI. Results of Stepwise Regression Analysis[a] for Case Herds in Two Ontario Counties

Variable Entered	Correlation Coefficient	Step at Which Variables Included	F Value to Enter Regression	Cumulative R-Squared
Test (diagnostic)	−0.388[b]	1	7.80[b]	0.15
Insemination method	0.137	2	3.27	0.21
Infected herds in 2 mile radius	0.182	3	2.26	0.25
Control herds within 2 miles	−0.170	4	0.94	0.27
Herd size	−0.044	5	1.35	0.29
History of previous reactor(s)	−0.098	6	0.62	0.30
Vaccination level	−0.259	7	0.65	0.31

Total $R^2 = 0.34$
[a]Dependent variable = Percent removals from herd. First seven independent variables entered into the regression are shown. Independent variables No. 1-13 Table III.
[b]Denotes significance at $p \lessapprox 0.05$

porting the view expressed by Schlotthauer (28).

2. The less the external exposure rating (Table III) the longer the time (no. tests and no. months) in quarantine. No obvious reason for this is apparent. It is possible that the differential rating of the factors comprising the exposure rating (Table IV) is incorrect since they were established on an intuitive basis from past experience.
3. The diagnostic test influenced the time required to become clean. Herds experiencing active infection, i.e. diagnosed on the basis of the abortion syndrome or MCT (44% of MCT suspect herds had clinical abortion or stillbirth history) required a longer time than those discovered by routine testing procedures, i.e. herds presenting minimal clinical evidence of infection (only 7% of case herds discovered on area testing and 17% of case herds found by traceback exhibited abortion or stillbirth). This concurs with clinical knowledge that describes abortion as the major disseminator of the infection (14).
4. Grade herds required longer to reach clean status. Perhaps this reflects the increased economic pressures exerted on the purebred owner to return to a *Brucella* free herd.
5. The vaccination level of cases was 16% and its variation among farms did not alter significantly the times required (although entered into the regression the F-value to enter was nonsignificant). Additional information on the effect of vaccination was provided by the results of the chi-square analysis displayed in Table XII which indicated that vaccination and reactor status were independent at the overall 16% level of vaccination. A twofold interpretation is possibe from this finding: (i) given the more liberal interpretation afforded vaccinates, such a vaccination level in infected herds does not appear to complicate HofA test procedures or (ii) no measurable protection is afforded individual cattle through vaccination when only 16% of susceptible cattle are vaccinated.

TABLE XII. The Association Between Vaccination and Serological Test Results at First Herd Test Subsequent to Quarantine

	Test Results			
	Negative	Questionable	Positive	
Vaccinates	112	5	7	124
Non-vaccinates	496	21	37	554
	608	26	44	678

Calculated $\chi^2 = 0.17$
Tabular χ^2 ($p = 0.05$) = 5.99, 2 d.f.

6. Natural service predisposed to longer quarantine periods than artificial insemination methods. This could reflect venereal transmission or factors related to natural service which in themselves prolong the quarantine period (e.g. loose housing). Rankin (26) and Bendixen (3) report that by itself insemination method does not strongly influence the natural history of brucellosis within a herd.

Stabling was the most significant estimator of the number of tests required to obtain a clean status, with loose housing tending to increase the number of tests. The greater potential for spread of the disease with this type of stabling (25) substantiates this finding.

The only variable of importance in predicting the percentage removals was the diagnostic test. The indication that active infection (i.e. those cases diagnosed by abortion or MCT as previously noted) predisposed to greater percentage losses is reasonable in consideration of the puerperal spread of the disease.

In this study we have identified factors which appear to heighten the risk of infection to a herd, to influence its spread within a herd and affect the logistics of returning to a clean herd status. The affect of these factors should be further investigated and constantly reassessed in the light of changing agricultural practices and departmental policies. It is our opinion that supplemented by information on control routinely collected HofA data if properly herds can be of considerable value for these purposes.

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